

- (c) culturing the producer cells in adherent mode in an adherent bioreactor having a fixed bed volume of at least about 5 liters whereby the producer cells produce the recombinant lentiviral vector; and then
- (d) harvesting recombinant lentiviral vector.
2. The method of claim 1, further comprising incubating the transfection solution for materially longer than 20 minutes, whereby the transfection solution does not form a DNA-PEI complex precipitate.
3. The method of claim 1, wherein the plasmid is present in an amount adequate to produce a PEI:plasmid DNA ratio of about 1:1.5.
4. The method of claim 1, wherein the transfection solution is at least about 20 liters in volume.
5. The method of claim 1, wherein the transfection is substantially complete before adding the transfection solution to the bioreactor.
6. The method of claim 1, wherein the line cells are added to the bioreactor before transfection is complete, and wherein the step of (b) adding line cells to the transfection solution further comprises circulating the transfection solution in the bioreactor until transfection is substantially complete.
7. The method of claim 1, wherein the plasmid DNA concentration in the transfection solution is at least about 300 nanograms of DNA per  $\text{cm}^2$ .
8. The method of claim 6, wherein the plasmid DNA concentration in the transfection solution is not more than about 400 nanograms of DNA per  $\text{cm}^2$ .
9. A method comprising:
- (a) combining PEI with plasmid DNA to make a transfection solution, and then measuring the formation of DNA-PEI complexes in the transfection solution using light scattering.
10. A method comprising:
- (a) combining PEI with plasmid DNA and cells in media in a bioreactor, the bioreactor configured to substantially cease the addition of  $\text{CO}_2$ , whereby the PEI does not substantially react with added  $\text{CO}_2$ .
11. A method comprising:
- (a) combining PEI with cells and with plasmid DNA in culture medium in a bioreactor, and
- (b) allowing the pH of the culture medium to fall naturally during or just after transfection, producing a slightly-acidic culture medium which prevents PEI-DNA complex precipitate formation.
12. The method of claim 2, further comprising stirring the transfection solution.
13. The method of claim 1, further comprising incubating the transfection solution until the DNA/PEI particle size decreases to be not more than about 80% of the maximum particle size.
14. The method of claim 9, further comprising:
- (b) adding line cells to the transfection solution whereby the plasmid transfects the line cells to make producer cells which produce a recombinant viral vector; and
- (c) culturing the producer cells in a volume of at least about 5 liters whereby the producer cells produce the recombinant viral vector; and then
- (d) harvesting recombinant viral vector.
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